I. Report Title: Development of hatchery technologies for snapper

Grantee:
G. Joan Holt (University of Texas Marine Science Institute)
D. Allen Davis (Auburn University)
Organization: University of Texas Marine Science Institute

Grant Number: NA07FD0173
Date: October 3, 2002

II. Abstract

This project examined the fatty acid requirements of yellowtail snapper larvae in order to expand our understanding of larval rearing requirements and advance commercial technologies for the production of yellowtail snapper fingerlings. Larval rearing trials have shown that feeding larvae rotifers and *Artemia* enriched with commercially available products such as Algamac 2000 or Aquagrow Advantage resulted in better growth than larvae fed the same feeds enriched with live algae (*Isochrysis galbana* and *Nannochloris occulata*). However, no significant differences in larval growth were found among treatments fed rotifers and *Artemia* enriched with Algamac 2000 and Aquagrow Advantage even though these enrichments resulted in significantly different levels of highly unsaturated fatty acids (HUFAs). Snapper larvae apparently require much higher levels of highly unsaturated fatty acids than other marine fish such as red drum (*Sciaenops ocellatus*). For example, *Isochrysis galbana* enriched rotifers are adequate for red drum larvae whereas yellowtail snapper require a more highly enriched diet. These results suggest that successful culture of this species is dependent upon supplying larvae with adequate levels of fatty acids such as docosahexaenoic acid (DHA), eicosapentaenoic acid (EPA) and arachidonic acid (ARA).

III. Executive Summary

This project examined the fatty acid requirements of yellowtail snapper larvae in order to expand our understanding of larval rearing requirements and

advance commercial technologies for the production of yellowtail snapper fingerlings. The results of this study suggest that the successful culture of yellowtail snapper is dependent upon enriching live feeds with high levels of HUFAs.

IV. Purpose

Ocean fishing, the largest source of fish production, is producing about 90 million metric tons annually and can no longer sustain the over fishing and pollution pressures man has placed on it. At the same time, the world's demand for seafood is rapidly increasing. In order to sustain, or in some cases rebuild, our fisheries we must not only use traditional management plans and restocking strategies, but we must also provide alternate sources of seafood (mariculture). Yellowtail snapper is one of several snapper species that are listed as "over fished" and displays positive potential for development in the mariculture industry. The specific goal of the project was to develop larval rearing techniques for the mass production of yellowtail snapper. Results from this project were expected to a) diversify the number of culture species available to the mariculturist b) expand our understanding of larval rearing requirements of yellowtail snapper and c) advance commercial technologies for the production of yellowtail snapper fingerlings.

V. Approach

Over the course of the study, two independent brood stock tanks of yellowtail snapper were induced to spawn multiple times via manipulations of temperature and photoperiod. A total of 9 larval rearing trials were conducted in order to gain insight into the fatty acid requirements of yellowtail snapper larvae and to refine mass production techniques for rearing these larvae. Nutritional trials were conducted in 150 L cone tanks equipped with an internal biological filter for maintaining water quality and mass production trials were conducted in a series of 12, 1,000 L larval rearing tanks set up to run either as flow through single pass or as a re-circulating system.

Eggs used in the larval rearing trials were collected from the brood stock tank the morning after spawning, set in a solution of 1 % formalin for 30 min and then transferred into the culture tanks. Eggs were enumerated volumetrically (1,000 eggs/ml) prior to stocking.

Larvae were initially fed rotifers on day 3 after hatching and were switched to enriched *Artemia* nauplii by day 18. Rotifers (*Brachionus plicatilis*) used in this study were obtained from a continuous culture raised on *Nannochloris occulata* and yeast. Each larval rearing trial included at least 2 different types of enrichments for rotifers and/or *Artemia*. The required number of rotifers was harvested the day before feeding, placed in a 20 l plastic container (300 rotifers/ml) and enriched with either live algae (*Nannochloris occulata* or *Isochrysis galbana*) or commercial enrichments (Algamac 2000 or Aquagrow Advantage). Rotifers were enriched twice over a 16 h period and maintained at 28.0 ppt and 26 C. *Artemia* cysts were incubated for 20 h at 28.0 ppt and 26 C at which time nauplii were separated from empty cysts and placed in a 20 l plastic container (50-100 nauplii/ml). The nauplii were enriched as described for rotifers. Enriched rotifers and *Artemia* were sampled at least twice during each trial for lipid analysis and dry weight determinations.

During each rearing trial, a subset of larvae was sampled from each rearing tank approximately 12 and 20 days after hatching for determination of larval growth and fatty acid analysis. Following collection, larvae were anesthetized with 0.1 % tricaine methanesulfonate (MS-222). Standard length measurements were made to the nearest 0.1 mm using a Wild Heerbrugg stereomicroscope, Summa Sketch III digitizing tablet (GTCO CalComp, Inc., Columbia, Maryland, USA) and Sigma Scan software (Jandel Corporation, San Rafael, California, USA). Larvae were then held at -80 C until fatty acid analysis could be performed. At the termination of each rearing trial, the number of larvae surviving in each treatment was determined.

A one-way ANOVA followed by a Tukey test for multiple comparisons of means was used to analyze differences in growth and survival during each rearing

trial. Values used in the analysis were transformed as necessary to meet the normality assumption of ANOVA. Sample means were entered into the analyses.

Project Management: This project was supervised by Drs. G. Joan Holt and D. Allen Davis. Maturation, spawning and larval rearing of yellowtail snapper was performed at the University of Texas Marine Science Institute by Cynthia Faulk (Research Scientist Associate) and Maotang Li (Research Scientist Assistant). Biochemical analyses were performed by Cynthia Faulk at the University of Texas Marine Science Institute and by Deborah R. Smith, an undergraduate who worked on the project and received training in lipid analyses at Auburn University.

VI. Findings

Fatty acid analysis of enriched rotifers and *Artemia* indicate that both commercial products used in this study, Algamac 2000 and Aquagrow Advantage, resulted in higher levels of highly unsaturated fatty acids (HUFAs) than either *Nannochloris occulata* or *Isochrysis galbana*. Larval rearing trials have shown that feeding larvae rotifers and *Artemia* enriched with these commercial products resulted in better growth than larvae fed the same feeds enriched with live algae. However, no significant differences in larval growth were found among treatments fed live feeds enriched with either Algamac 2000 or Aquagrow Advantage even though these enrichments resulted in significantly different levels of HUFAs including docosahexaenoic acid (DHA), eicosapentaenoic acid (EPA) and arachidonic acid (ARA). These results suggest that yellowtail snapper larvae have a high requirement of HUFAs especially DHA.

Previous yellowtail snapper rearing trials conducted in our laboratory resulted in poor survival near day 18 post-hatch due to an apparent stress response which is especially severe during periods of handling. Similar responses have been reported with mahimahi *Coryphaena hippurus* in which larvae had been subjected to a stress test following growth trials using several diets (Kraul et al. 1993). Mahimahi larvae fed diets with reduced levels of docosahexaenoic acid (DHA)

had less resistance to stress shock. One goal of this study was to decrease the frequency and/or severity of this response by providing increased amounts of HUFAs through the diet. Although increasing the amount of HUFAs in the live feeds increased the growth rate of yellowtail snapper no differences were found in overall survival or resistance to stress. It is widely accepted that *Artemia* nauplii are an inadequate source of nutrition for marine fish larvae even when enriched with high levels of HUFAs because they rapidly retroconvert longer chain fatty acids to shorter chain fatty acids. In this study, the fatty acid profile of enriched *Artemia* was substantially less than that of the rotifers and that of larval dry feeds typically used to rear marine fish larvae. It is possible that the enriched Artemia did not provide an adequate supply of HUFAs to the larvae which resulted in the occurrence of the stress response and low overall survival rates. Further research is needed to develop an early weaning protocol for this species in which larvae are switched from *Artemia* to a formulated diet prior to the age at which the stress response is most prevalent (approximately day 18 post-hatch).

Kraul, S., K. Brittain, R. Cantrell, T. Nagao, H. Ako, A. Ogasawara and H. Kitagawa. 1993. Nutritional factors affecting stress resistance in the larval mahimahi *Corphaena hippurus*. Journal of the World Aquaculture Society 24: 186-193.

VII. Evaluation

Results obtained from the 9 larval rearing trials conducted over the course of this study allowed us to meet our objectives by enhancing our knowledge of the fatty acid requirements of the larvae and determining the need for enrichment of rotifers, *Artemia* or both. Additional research is still needed to define an optimal diet and early weaning strategy for rearing yellowtail snapper larvae.

Results of this study will be made available to interested parties via presentations at scientific meetings and publication of results in scientific journals.

Oral presentation:

World Aquaculture Society 2003

Presenter: D. Allen Davis

Title: Yellowtail snapper culture: evaluation of live food enrichments

Manuscript in preparation:

Authors: Cynthia K Faulk, G. Joan Holt and D. Allen Davis

Tentative title: Evaluation of fatty acid enrichment of live food for yellowtail snapper larvae